REGULAR ARTICLE

blood advances

Cerebral hemodynamic changes after haploidentical hematopoietic stem cell transplant in adults with sickle cell disease

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Key Points

- Adults with SCD had improved cerebral hemodynamics after SCT and were not different compared with adult controls.
- Central nervous system events are rare after haploidentical SCT.

Preliminary evidence from a series of 4 adults with sickle cell disease (SCD) suggests that hematopoietic stem cell transplant (HSCT) improves cerebral hemodynamics. HSCT largely normalizes cerebral hemodynamics in children with SCD. We tested the hypothesis in adults with SCD that cerebral blood flow (CBF), oxygen extraction fraction (OEF), and cerebral metabolic rate of oxygen ($CMRO_2$) measured using magnetic resonance imaging, normalized to healthy values, comparing measurements from ~1 month before to 12 to 24 months after HSCT (n = 11; age, 33.3 ± 8.9 years; 389 ± 150 days after HSCT) with age-, race- and sex-matched values from healthy adults without sickle trait (n = 28; age, 30.2 ± 5.6 years). Before transplant, 7 patients had neurological indications for transplant (eg, overt stroke) and 4 had nonneurological reasons for haploidentical bone marrow transplant (haplo-BMT). All received haplo-BMT from first-degree relatives (parent, sibling, or child donor) with reduced-intensity preparation and maintained engraftment. Before transplant, CBF was elevated (CBF, 69.11 ± 24.7 mL/100 g/min) compared with that of controls (P = .004). Mean CBF declined significantly after haplo-BMT (posttransplant CBF, 48.2 ± 13.9 mL/100 g/min; P = .003). OEF was not different from that of controls at baseline and did not change significantly after haplo-BMT (pretransplant, 43.1 ± 6.7%; posttransplant, 39.6 ± 7.0%; P = .34). After transplant, CBF and OEF were not significantly different from controls (CBF, 48.2 ± 13.4 mL/100 g/min; P = .78; and OEF, 39.6 ± 7.0%; P > .99). $CMRO_2$ did not change significantly after haplo-BMT (pretransplant, 3.18 ± 0.87 mL O_2/100 g/min; posttransplant, 2.95 ± 0.83; P = .56). Major complications of haplo-BMT included 1 infection-related death and 1 severe chronic graft-versus-host disease. Haplo-BMT in adults with SCD reduces CBF to that of control values and maintains OEF and $CMRO_2$ on average at levels observed in healthy adult controls. The trial was registered at www.clinicaltrials.gov as #NCT01850108.

Introduction

Adults with sickle cell disease (SCD) are at high risk of overt stroke and silent cerebral infarction (SCI).[1] Brain injury progresses throughout the lifespan,[2] resulting in cognitive symptoms, decreased quality of life, and long-term disability.[3-6] Currently, the mainstays of disease-modifying therapy include

Submitted 12 May 2023; accepted 16 October 2023; prepublished online on *Blood Advances* First Edition 26 October 2023; final version published online 26 January 2024. https://doi.org/10.1182/bloodadvances.2023010717.

Data are available on request from the corresponding author, Lori C. Jordan (lori.jordan@vumc.org).

The full-text version of this article contains a data supplement.

hydroxyurea and regular blood transfusions, although unfortunately, new cerebral infarcts occur in some patients despite interventions.[7] Curative therapy such as hematopoietic stem cell transplant (HSCT) using haploidentical bone marrow transplant (haplo-BMT) is appealing because haplo-BMT should improve cerebral hemodynamics, reduce the risk of new infarcts,[8,9] and prevent progressive cerebral vasculopathy[10] with better availability than standard BMT.[11] However, other than a case series of 4 adults with improved cerebral hemodynamics after haplo-BMT,[12] this has not yet been confirmed in adults with SCD. Mechanisms of cerebral infarcts in SCD may be due to inadequate cerebral blood flow (CBF) from anemia, hemoglobin S (HbS), and, in some cases, arterial steno-occlusion. Currently, our understanding of how cerebral hemodynamics related to stroke risk change after haplo-BMT is limited.

As first demonstrated using gold-standard ^{15}O positron emission tomography (PET) hemodynamic imaging,[13] CBF (mL/100 g per minute)[14-16] in adults and children with SCD is frequently elevated to maintain adequate oxygen delivery to a healthy cerebral metabolic rate of oxygen ($CMRO_2$).[13] CBF increases primarily due to relaxation of smooth muscle lining cerebral arterioles, and the oxygen extraction fraction (OEF) may increase, in the presence of preserved $CMRO_2$, if elevated CBF is insufficient to maintain adequate oxygen delivery. Whole-brain OEF is elevated in many children and adults with SCD and correlates inversely with total Hb.[14,17,18] Ford et al have shown that in children with SCD, OEF is highest in the brain regions with the lowest CBF, which are also the regions at the greatest risk of SCIs.[19] A related complication is that higher blood flow velocities may result in faster red cell and plasma transit through capillaries, decreasing the capillary transit time and ability to offload oxygen into surrounding tissues, that is, capillary shunting.[20] Shunting effects have been observed as dural venous sinus hyperintensities on the commonly performed pseudocontinuous arterial spin labeling (pCASL) magnetic resonance imaging (MRI) method and are more prevalent in patients with SCD compared with age- and race-matched controls.[21] CBF, OEF, and capillary shunting are potential biomarkers of stroke risk that may be assessed via noninvasive MRI methods.

Cerebral hemodynamic measures have been shown to improve with SCD stroke prevention therapies including oral hydroxyurea[17,22] and chronic blood transfusion.[16,17] Logically, haplo-BMT should improve cerebral hemodynamics more than blood transfusions, although reports on using these as biomarkers for measuring improvements in cerebrovascular health after haplo-BMT are limited. Recently, normalization of cerebral hemodynamics has been reported by Hulbert et al in children with SCD after HSCT using noninvasive brain MRI methods.[23] Significant improvements in both OEF and CBF were seen in 10 children who underwent HSCT compared with children with SCD receiving regular blood transfusions.[23] However, children with SCD have been shown to have a better cerebral hemodynamic response to blood transfusions than adults[16] and have previously tolerated HSCT better than adults.[24]

Historically, adults with SCD and myeloablative transplants have also had lower survival rates and event-free survival than children.[25] Availability of related sibling donors is limited to ~6% of affected families;[26] ~18% of patients with SCD will have a fully HLA-matched unrelated donor in the worldwide registry.[27] These limitations have resulted in the development of reduced-intensity, related, haploidentical (or half-match), transplant protocols that have expanded the donor pool with first-, second-, and third-degree relatives as possible donor sources.[11,28] We previously reported on outcomes of 4 adults with SCD who underwent haplo-BMT including 1 with syndromic moyamoya and new SCI during the course of the haplo-BMT.[12] Here, we report a larger cohort of 11 adults with SCD and haplo-BMT who received multicontrast anatomical and functional neuroimaging within 1 month before and 12 to 24 months after haplo-BMT and compare them with age-, sex-, and race-matched control imaging metrics. Although new or progressive infarcts after haplo-BMT for SCD are uncommon, insight regarding cerebral hemodynamic changes in adults after transplantation may provide useful future biomarkers for understanding who may be most at risk of future stroke in patients without transplant. Additionally, transplant procedures vary across different sites, so, multiple reports across various ages and populations will allow for the best understanding of neurological outcomes and how cerebral hemodynamics change in this population. We tested the physiological hypothesis that CBF reduces after haplo-BMT, approaching that of healthy control values. Secondary hypotheses are that the OEF change is related to the pre–haplo-BMT OEF and that imaging markers of capillary shunting reduce following haplo-BMT.

Methods

Participants provided informed, written consent for this prospective study. All protocols were approved by the Vanderbilt University Medical Center Institutional Review Board (study 211317). Participants underwent neuroimaging before and after clinically indicated haplo-BMT. Inclusion criteria included adults aged ≥18 years with SCD undergoing haplo-BMT following the established BMT protocol 12108 (NCT01850108).[29] All patients received pretransplant conditioning with thymoglobulin, thiotepa, fludarabine, and low dose total body irradiation (200 cGy), bone marrow as graft source, and graft-versus-host disease prophylaxis (supplemental Figure 1).

Pretransplant and posttransplant MRI and magnetic resonance angiography (MRA) of the brain were obtained under the haplo-BMT protocol, and cerebral hemodynamic MRI sequences were added using separate funding. Exclusion criteria included neurological conditions unrelated to SCD such as congenital brain malformations, history of traumatic brain injury, or inability to tolerate MRI. Age-, sex-, and race-matched controls (n = 28; age = 30.2 ± 5.6 years) without sickle trait with identical exclusion criteria were enrolled for comparison at ~2:1 ratio. Age matching was performed by matching year of birth ±3 years. Participants underwent standardized neurological examination by a board certified neurologist. Phlebotomy was performed within 7 days of MRI to record Hb and hematocrit values, and high-performance liquid chromatography was used to assess the percentage of HbS. Adult controls underwent venipuncture and the same MRI protocol once to assess hematologic and hemodynamic values for comparison with the SCD population.

MRI

Participants completed a noncontrasted brain MRI and MRA of the head and neck (3.0T Philips, Best, The Netherlands) within 1 month before and with a goal of 12 months after haplo-BMT per

protocol. Arterial oxygen saturation values were obtained using pulse oximetry concurrent with imaging.

Time-of-flight MRA of the head and neck assessed for intracranial and cervical vasculopathy; MRI of the brain, including axial and coronal T_2-weighted fluid-attenuated inversion recovery, axial diffusion-weighted imaging, and T_2-weighted and 3D T_1-weighted imaging, evaluated for new or progressive cerebral infarcts. Infarcts were judged to be silent based on history and detailed neurological examination. For OEF measurement, T_2-relaxation-under-spin-tagging (TRUST) data were acquired twice per session (spatial resolution = 3.4 × 3.4 × 5 mm; τ-CPMG = 10 ms; eTE = 0, 40, 80, and 160 ms; TR/TE = 1978/3.6 ms; averages = 3) using a common protocol previously validated and evaluated for reproducibility.[14,29,30] Control and venous-labeled (transfer insensitive labeling technique) TRUST images were acquired from a slice containing the superior sagittal sinus, ~20 mm superior to the torcula.[14,15,21,31] CBF and assessment of venous hyperintense signal were measured with a multislice 2D pCASL sequence (postlabeling delay = 1900 ms; spatial resolution = 3.0 × 3.0 × 7.0 mm) with 20 blocks of alternating acquisitions with and without spin labeling and without background suppression.

Analysis

Two neuroradiologists, blinded to patient status and clinical course, independently evaluated for cerebral infarcts and vasculopathy; disagreements were reconciled by consensus discussion. Infarct was defined as T_2-weighted fluid-attenuated inversion recovery hyperintensity visible on 2 imaging planes and ≥3 mm in 1 plane.[32] Vasculopathy was defined as stenosis of a major cervical (internal carotid or vertebral artery) or intracranial (basilar or first segment of the middle cerebral artery, anterior cerebral artery, or posterior cerebral artery) vessel >50% using warfarin-aspirin symptomatic intracranial disease criteria.[33]

For OEF quantification, TRUST data were pair-wise subtracted and venous T_2 values within the superior sagittal sinus were converted to venous oxygen saturation (Y_v) using previously characterized human blood calibration curves.[34] Y_v and arterial oxygenation saturation (Y_a) were used to calculate OEF as the fractional difference of blood oxygenation in the arteries and veins (OEF = 100% × $[Y_a - Y_v]/Y_a$). CBF was calculated[35] using blood arrival time measures recently measured from adults with SCD and in controls.[21] The subject-specific arterial blood T_1, calculated using measured hematocrit, was also included in the model.[36]

Dural venous sinus hyperintense signal on pCASL images has been proposed as a marker of rapid red cell and plasma transit through intracranial vasculature because these hyperintensities correlate directly with macrovascular flow velocities,[37] are inversely related to oxygen extraction,[31] and reduce after improvements in blood oxygen content secondary to red cell exchange transfusions.[38] Venous sinus signal was quantified using the pCASL data, whereby a circular region of interest with cross-sectional area 40 mm^2 was evaluated at the approximate location of the parieto-occipital sulcus, and calibrated mean venous flow signal within the region of interest was extracted from quantified CBF maps, as described previously.[38] Of note, the quantified CBF maps were used for this determination such that normalized luminal signal could be compared across participants and sessions without interscan concerns related to differences in signal scaling or scanner gain.

$CMRO_2$ was calculated based on the Fick principle (using $CMRO_2$ = CBF × OEF × oxygen content), in which oxygen content is approximated as the product of the bound oxygen (1.34 mL O_2 per g Hb) and the Y_a.[34,39] Note that $CMRO_2$ reported in the results section is converted to the common units (mL O_2/100 g tissue per minute).

All participants underwent the full imaging protocol, but sometimes, because of artifacts or motion during an imaging sequence, the data were not always usable. This was the case for OEF and $CMRO_2$ data from 1 participant.

Statistical methods

Participant characteristics are described by mean and standard deviation for continuous measures and by number and percent for categorical measures. Data are presented as line graphs, box-and-whisker plots, and/or in violin plots.

To test the hypotheses that CBF values are (1) elevated in SCD compared with that of controls and (2) reduced to control levels after haplo-BMT, we first applied a 2-tailed Wilcoxon signed-rank test. We then compared pretransplant and posttransplant CBF values of participants with SCD with those of controls using a Kruskal-Wallis test. Additionally, we used the pCASL CBF values to test the hypothesis that signal in the superior sagittal sinus of patients with SCD would significantly decrease after haplo-BMT and assessed these changes using a 2-tailed Wilcoxon signed-rank test.

To test the secondary hypotheses that OEF values are (1) elevated in SCD compared with those of controls and (2) reduced to control levels after haplo-BMT, we first applied a 2-tailed Wilcoxon signed-rank test. Next, we assessed how baseline OEF percentage relates to overall change in OEF also using a 2-tailed Wilcoxon test. We then compared SCD pretransplant and posttransplant OEF values with control values using a Kruskal-Wallis test. This method was also used to test OEF models in the supplemental Material.

Lastly, we tested the hypothesis that $CMRO_2$ values are (1) elevated in SCD compared with those of controls and (2) reduced to control levels after haplo-BMT. We applied a 2-tailed Wilcoxon signed-rank test. We then compared pretransplant and posttransplant $CMRO_2$ values of participants with SCD with those of controls using a Kruskal-Wallis test.

For all analyses, significance was defined as 2-sided P value < .05, corrected for multiple comparisons if necessary. For statistical testing using Wilcoxon test, we used Bonferroni corrections, and for Kruskal-Wallis tests, Dunn multiple comparisons testing was applied.

Results

Demographics

Pre– and post–haplo-BMT cerebral hemodynamic imaging was obtained in 11 adults with SCD (Table 1; age = 33.3 ± 8.9 years), including 6 females (55%), 8 with HbSS, 2 HbSC, and 1 HbS β^+ thalassemia. Before transplantation, 5 had overt stroke, of whom 4 had SCD-related moyamoya syndrome, 2 had silent cerebral infarcts, and 4 had nonneurological reasons for haplo-BMT. All patients achieved full engraftment (100% myeloid donor cells) at the time of follow-up imaging. Note that 4 participants were included in a prior publication.[12] Only 3 of 11 participants (27%)

Table 1. Patient-level data for adults with SCD and HSCT

ID	Genotype	Transplant indications	Age (y), sex	DMT	Haplo-BMT Hb (g/dL) (before, after)	Haplo-BMT HbS% (before, after)	Stem cell donor relation, genotype	Myeloid engraftment (% donor)	Change in mRS and mRS scores before/after transplant	Transplant complications
1*	HbSS	ACS and osteomyelitis	20.8, M	HU	6.7, 12.9	80.5, 32.1	Sibling, HbAS	100%	None, 0/0	N/A
2*	HbSS	Overt stroke and moyamoya	34.4, M	HU†	6.7, 14.2	86.9, 32.8	Sibling, HbAS	100%	None, 3/3	HHV6 encephalitis treated with Foscarnet.
3*	HbSS	SCI, ACS, VOC, and iron overload	39.9, F	HU	5.2, 10.9	60.6, 31.6	Sibling, HbAS	100%	None, 0/0	N/A
4	HbSS	Overt stroke, moyamoya, and ACS	24.6, M	CBT	9.4, 13.1	44.4, 34.8	Parent, HbAS	100%	None, 1/1	First transplant did not engraft. Second transplant engrafted.
5*	HbSS	Overt stroke and moyamoya	39.5, F	HU + CBT	7.4, 11.2	18.0, 0.0	Sibling, HbAA	100%	None, 1/1‡	New SCI at 8.5 months after transplant (MRI timing for participant convenience, not symptoms) and died of COVID-19 3.5 y after transplant.
6	HbSS	Overt stroke and moyamoya	25.9, F	CBT	10.4, 8.1	35.1, 0.0	Sibling, HbAA	100%	None, 3/3	Major ABO-incompatibility with transplant resulting in pure red cell aplasia. Eventually able to treat with monoclonal antibody daratumumab at 4 doses; current Hb level at 12.9 g/dL.
7	HbSS	SCI, ACS, and pulmonary HTN	43.7, F	HU + CBT	10.4, 14.3	15.2, 38.8	Sibling, HbAS	100%	None, 0/0	N/A
8	HbSThal+	VOC requiring chronic transfusions, and hyperhemolytic phenotype	22.4, M	CBT	10.2, 19.2	34.9, 38.2	Sibling, HbAS	100%	None, 0/0	N/A
9	HbSC	Overt stroke, SCI, ACS, and systemic lupus	43.7, F	CBT	11.1, 13.1	31.3, 0.0	Sibling, HbAA	100%	Improved, 1/0	Significant ocular GVHD.
10	HbSC	ACS, pulmonary embolus, and retinal hemorrhage	41.8, M	HU	10.4, 12.9	12.9, 33.4	Child, HbAS	100%	None, 0/0	N/A
11	HbSS	ACS and AVN of hips with hip replacements	29.6, F	HU	8.4, 15.0	73.8, 39.8	Sibling, HbAS	100%	None, 0/0	N/A

ACS, acute chest syndrome; AVN, avascular necrosis; CBT, chronic blood transfusion therapy; CSVT, cerebral sinovenous thrombosis; DMT, disease-modifying therapy; DVT, deep vein thrombosis; GVHD, graft-versus-host disease; HHV-6, human herpes virus-6; HTN, hypertension; HU, hydroxyurea; SCI, silent cerebral infarct; TIA, transient ischemic attack; VOC, vaso-occlusive crisis.

*Previously reported.[12]

†CBT declined.

‡mRS at the time of neuroimaging, before death from infection.

had HbAA first-degree relative donors, 8 (73%) had HbAS donors and, therefore, posttransplant HbS levels reflected the trait status of their respective donors.

Hematology

In participants with SCD (n = 11), Hb increased from 8.8 ± 2.0 g/dL before transplant to 12.9 ± 2.9 g/dL after transplant (P = .005; Table 2). Hematocrit increased from 25.4% ± 5.6% to 38.3% ± 8.1% after haplo-BMT (P = .005). HbS percentage reduced from 44.9% ± 26.7% to 25.6% ± 16.7% (P = .04). In 1 participant, Hb did not increase during the first year after transplantation because of an ABO-incompatible donor transplant resulting in pure red cell aplasia. Non-SCD control participants had Hb of 13.4 ± 1.7 g/dL and hematocrit of 40.8% ± 4.0% (Table 2).

Cerebral hemodynamics

Pretransplant and posttransplant CBF assessment was available for all 11 participants. OEF values were not available for 1 participant because of imaging artifacts that rendered OEF uninterpretable (supplemental Table 1). Posttransplant MRIs were completed in a mean of 389 ± 150 days after haplo-BMT.

Healthy control (n = 28) CBF mean values were 47.5 ± 4.4 mL/100 g per minute; in adults with SCD, pretransplant mean CBF was elevated relative to that of control participants (P = .004) and significantly decreased from before transplant (CBF = 69.1 ± 24.8 mL/100 g per minute) to after transplant (CBF = 48.2 ± 13.9 mL/100 g per minute; P = .003; Figure 1A). Mean gray matter CBF maps are presented in Figure 2. After haplo-BMT, adults with SCD did not show significant CBF differences compared with control participants (P = .78; Figure 1B).

In adults with SCD (n = 11), mean signal in the superior sagittal sinus, quantified in units of mL/100 g per minute to allow for comparison with CBF signal, significantly decreased from before transplant (54.0 ± 28.7 mL/100 g per minute) to after transplant (18.9 ± 7.25 mL/100 g per minute; $P < .001$; Figure 3A), suggesting less capillary shunting.

In adults with SCD with OEF data (n = 10), mean OEF before transplant (OEF = 43.1% ± 6.7%) did not significantly change after haplo-BMT (OEF = 39.6% ± 7.0%; P = .28; Figure 4A). Patients with SCD who have a baseline OEF percentage that is greater than 1 standard deviation above the average seen in controls (OEF > 42%)[16] tend to show decreased OEF, whereas those in a more normal OEF range at baseline do not show decreases in OEF after transplantation (Figure 5). Compared with OEF in non-SCD controls (37.7% ± 6.1%), adults with SCD do not show significant differences in their OEF either before (P = .13) or after haplo-BMT ($P > .99$; Figure 4B). Relationships between before transplantation and OEF change after transplantation were directionally similar regardless of the TRUST calibration model used, and no model demonstrated a mean significant change in OEF before transplant vs after transplant. Results from other models are provided in supplemental Material.

In adults with SCD (n = 10), mean $CMRO_2$ before transplantation ($CMRO_2$ = 3.2 ± 0.9 mL O_2/100 g per minute) did not significantly change after transplantation ($CMRO_2$ = 2.9 ± 0.8 mL/100 g per minute) (Figure 6A). Patients with SCD have similar $CMRO_2$ values compared with controls ($CMRO_2$ = 3.0 ± 0.5 mL O_2/100 g per minute) and do not show significant differences either before (P = .37) or after haplo-BMT ($P > .99$; Figure 6B).

Major complications after transplantation in patients included serious viral infections in 2 patients (human herpes virus 6 encephalitis and human herpes virus 6 bacteremia), Epstein-Barr virus and cytomegalovirus reactivation in 1 patient each, and chronic graft-versus-host disease in 1 participant (supplemental Table 2), similar to other published haplo-BMT studies.[40,41] One participant with major ABO-incompatible transplantation developed pure red cell aplasia with severe anemia, and transfusion dependence was still present at the time of 1-year posttransplant MRI, so cerebral hemodynamics were not improved; their red cell aplasia recently resolved with treatment with daratumumab (a human monoclonal antibody that binds to CD38). One participant died of severe acute respiratory syndrome coronavirus 2 infection ~3.5 years after transplantation; this same participant (ID number 5) had moyamoya syndrome and was previously reported as having a new SCI seen on MRI at 8.5 months after transplantation[42]; no other participants had progression of SCI or new stroke. At baseline, 4 of the 11 participants (Table 1) had moyamoya-type severe cerebral vasculopathy, and in posttransplant follow-up, no change in intracranial stenosis degree by MRA or change in neurological examination was observed. To assess functional outcome, the modified Rankin scale (mRS)[43] was scored at the time of neuroimaging and neurological examination before and after transplantation; mRS was improved from 1 to 0 in 1 participant and unchanged in 10 of 11 participants at the time of neurological examination.

Table 2. Hematologic and cerebral hemodynamic parameters before and after transplant

	Healthy Controls	Pre–haplo-BMT	Post–haplo-BMT*	P†
Hb, g/dL	13.4 ± 1.7	8.8 ± 2.0	12.9 ± 2.9	<.001; .81; .005
Hematocrit, %	40.8 ± 4.0	25.5 ± 5.1	37.9 ± 7.7	<.001; .14; <.001
HbS, %	0.0 ± 0.0	41.7 ± 25.0	25.7 ± 17.1	<.001; <.001; .042
CBF, mL/100 g per minute	47.5 ± 4.4	67.7 ± 22.5	47.8 ± 12.5	.004; .78; .003
OEF, %	37.7 ± 6.1	43.4 ± 0.07	41.4 ± 0.07	.13; >.99; .34
$CMRO_2$, mL O_2/100 g per minute	3.0 ± 0.5	3.3 ± 0.9	3.0 ± 0.8	.37; >.99; .56

Values in the first 3 columns are shown as the mean ± the standard deviation.

*All values in this column are at the time of posttransplant MRI, 389 ± 150 days after transplant.

†Statistical results show P value comparisons between healthy control and pretransplant values, followed by comparisons between healthy controls and posttransplant values, followed by comparisons between pretransplant and posttransplant (haplo-BMT) values.

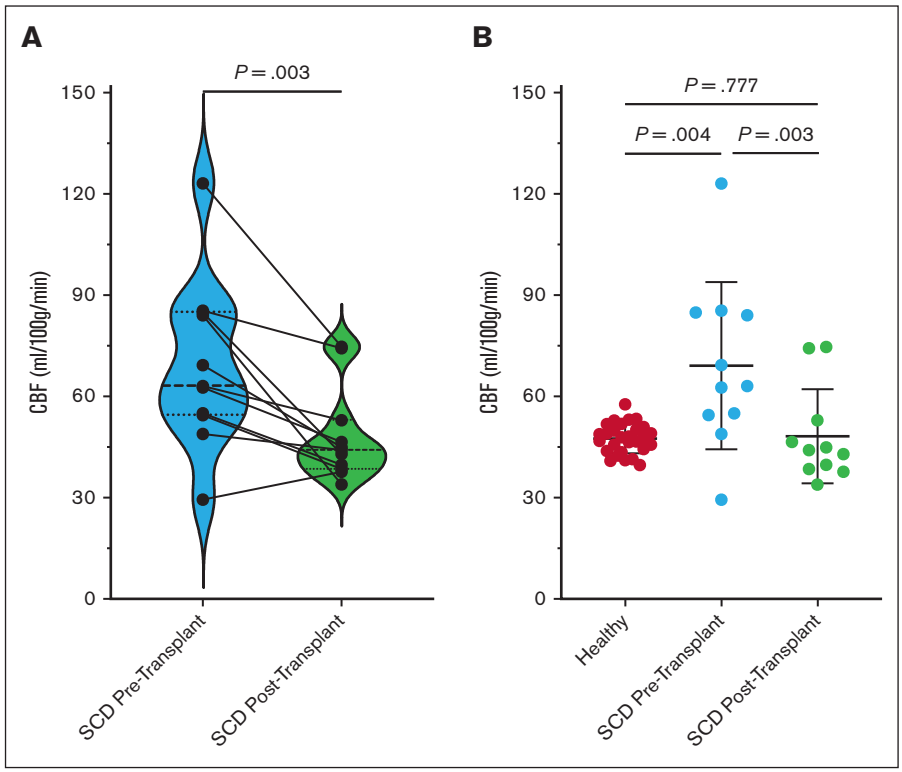

Figure 1. CBF changes before and after transplant and in relation to those in controls. (A) CBF changes in adults with SCD before and after HSCT. CBF significantly decreases after HSCT ($P = .003$). Individual participant changes are shown overlayed on the symbol and line graph, whereas group data are shown in violin plots, with the bolded dashed line showing the median and smaller dashed lines showing the upper and lower quartiles for participants before (blue plot) and after transplant (green plot). (B) Box-and-whisker plots of CBF shown in healthy adults (left; red dots) and in adults with SCD before transplant (middle; blue dots) and 1 year after transplant (right; green dots). CBF is significantly elevated in patients with SCD before transplant ($P = .004$) and reduces to levels similar to healthy adults after transplant ($P = .777$).

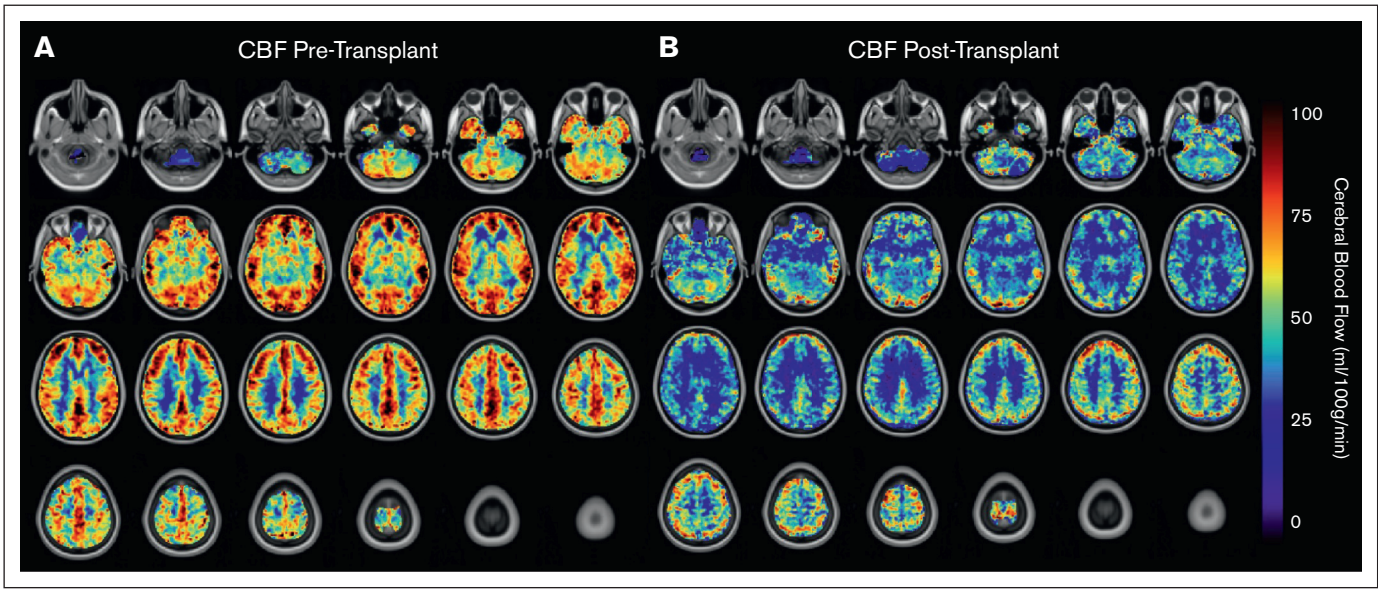

Figure 2. CBF changes before and after transplant shown in axial blood flow maps in MNI space. Ascending axial slices of CBF (mL/100 g per minute) maps averaged across participants with SCD before (A) and after (B) HSCT. Maps are shown overlayed on standard (Montreal Neurological Institute) space anatomical T1 images.

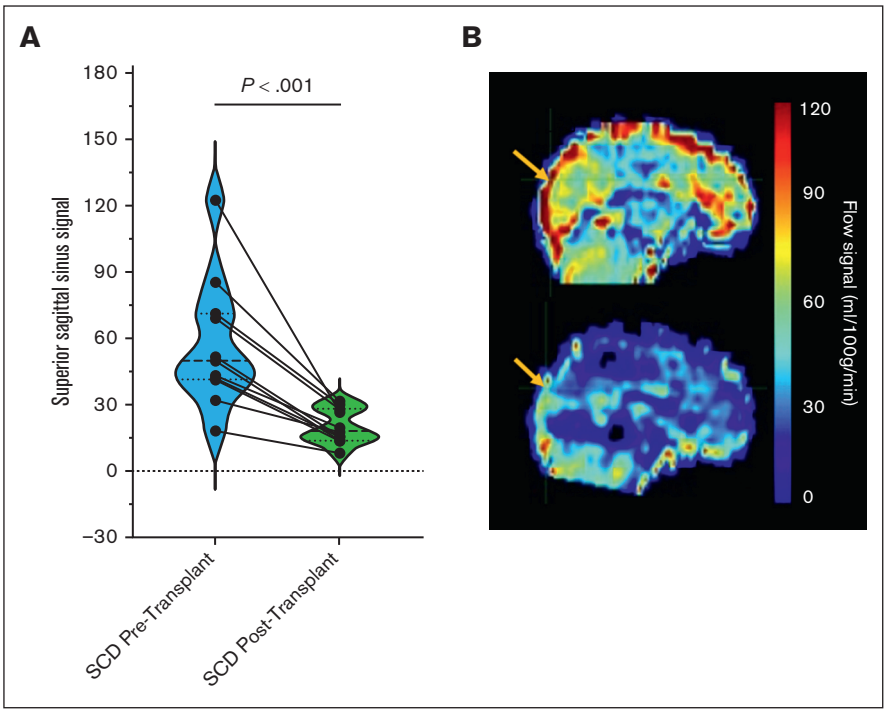

Figure 3. Superior sagittal sinus (SSS) flow changes from before to after transplant shown in graphical display and blood flow maps. (A) Individual subject changes are shown overlayed on the symbol and line graph, while group data are shown in violin plots with the bolded dashed line showing the median and smaller dashed lines showing the upper and lower quartiles for subjects pretransplant (blue plot) and posttransplant (green plot). (B) Signal change in the SSS quantified and shown in sagittal views of mean CBF shunting effect before transplant (top) and following transplant (bottom). Blood flow maps have been registered to standard (Montreal Neurological Institute) space; yellow arrows indicate the area of the sagittal sinus surveyed.

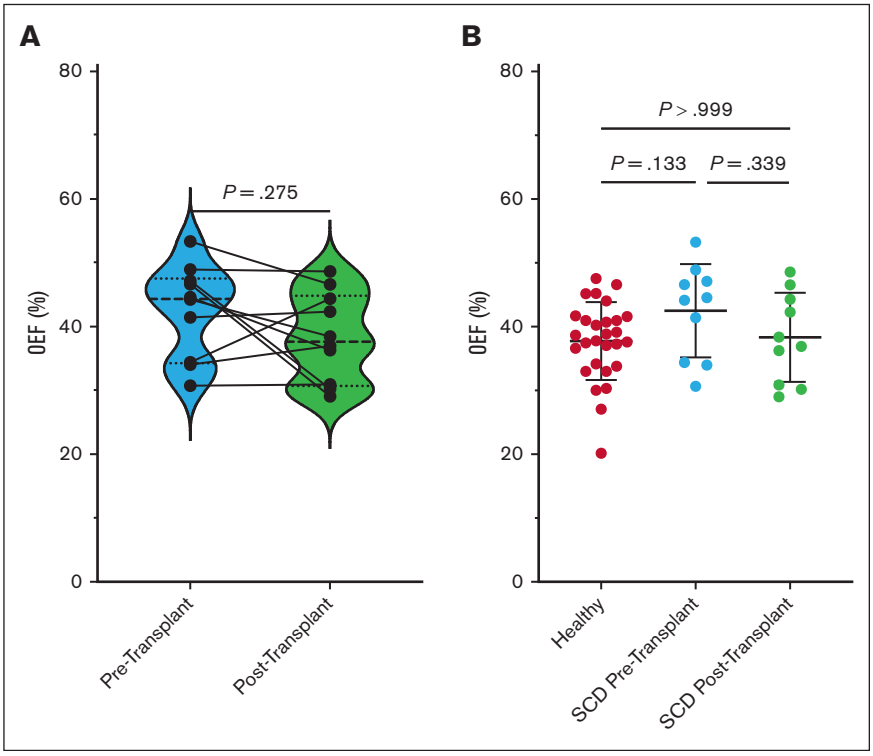

Figure 4. OEF changes from before to after transplant and compared with healthy controls. (A) OEF does not significantly change in adults with SCD before and after HSCT ($P = .28$). Individual participant changes are shown overlayed on the symbol and line graph, whereas group data are shown in violin plots, with the bolded dashed line showing the median and smaller dashed lines showing the upper and lower quartiles for participants before (blue plot) and after transplant (green plot). (B) Box-and-whisker plots of OEF shown in healthy adults (left; red dots) and in adults with SCD before transplant (middle; blue dots) and after transplant (right; green dots). OEF in patients with SCD is similar to values found in healthy adults both before ($P = .13$) and after transplant ($P = .34$).

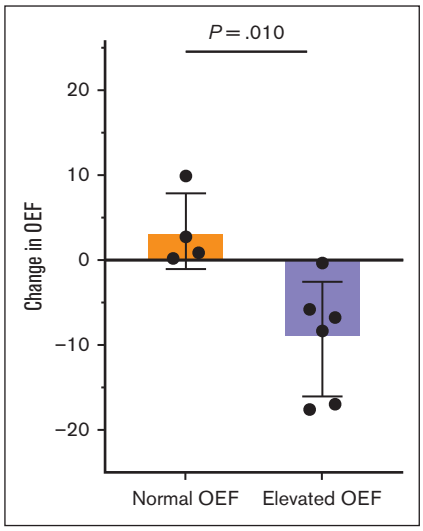

Figure 5. OEF changes relative to OEF baseline. OEF changes after transplant are significantly different in patients with SCD with a normal OEF (baseline OEF < 42%) than those with an elevated OEF at baseline (OEF > 42%; $P = .010$). Data are shown on bar graphs with the mean and standard deviation and individual participant data as dot overlay.

Discussion

In this cohort of adults with severe SCD, we demonstrate that CBF improves in most participants by reducing to near control levels after successful haplo-BMT. Cerebral hemodynamic compensation for anemia and reduced oxygen carrying capacity related to HbS are significantly improved as seen by the reduction in CBF values. OEF values do not change significantly on average; however, we observed a general inverse relationship between the pre–haplo-BMT OEF and the OEF change, suggesting that those with highest OEF before transplantation are likely to have the largest reduction in OEF after transplantation. We observed no difference in the $CMRO_2$ of patients before or after haplo-BMT, compared with that of controls.

After haplo-BMT, we find that average Hb levels significantly increase (Table 2; $P = .005$) and the average HbS percentage reduces by ~20%, reflecting the trait status of the donor (Table 2). In this study, we enrolled patients with existing severe cerebral vasculopathy (eg, moyamoya syndrome related to SCD), and these patients remain at risk of stroke and SCI on this basis.[42] Fluid and blood pressure shifts during the transplant period logically may put these patients at greater risk of stroke or SCI during the procedure. However, it is reassuring that of our 4 participants with moyamoya syndrome, only 1 had a new SCI visible on posttransplant MRI, and no participants had a new overt stroke. Neurological function as assessed by detailed neurological examination and mRS was unchanged in most and mildly improved in 1 participant. As an exploratory analysis, we assessed whether CBF changes were different between patients who had neurological indications for transplant vs those with other indications (supplemental Figure 2A) as well as those solely on hydroxyurea therapy vs only transfusion therapy before transplant (supplemental Figure 2B), although there were no significant differences between groups for either assessment. The stable increase in Hb and reduced endothelial injury after reductions in HbS related to haplo-BMT should be beneficial long term for individuals with SCD and severe cerebral vasculopathy;

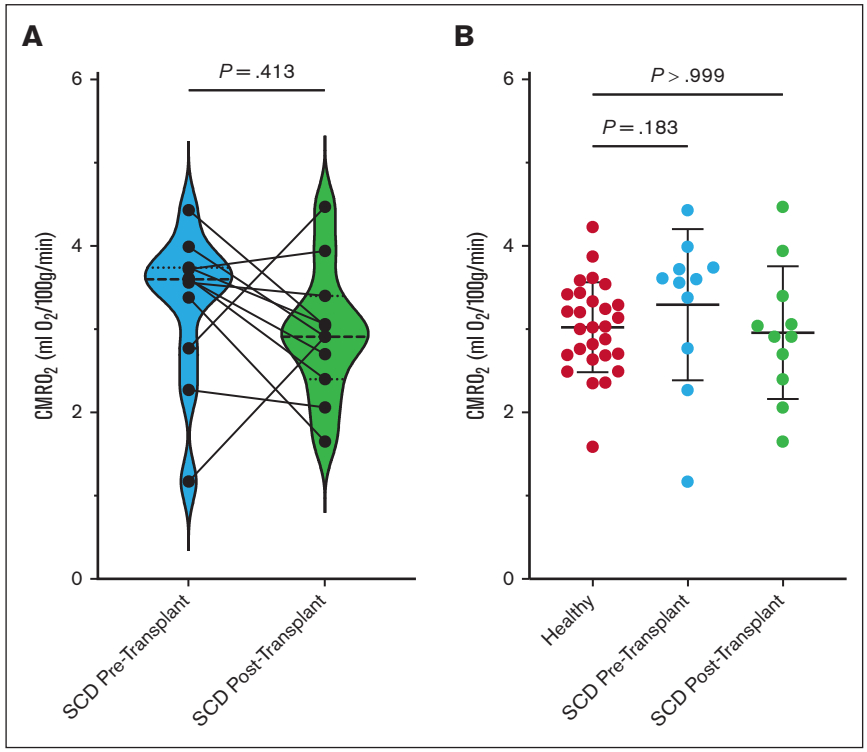

Figure 6. $CMRO_2$ changes before and after transplant and compared with healthy controls. (A) $CMRO_2$ changes in adults with SCD before and after HSCT (haplo-BMT). Individual participant changes are shown overlayed on the symbol and line graph, whereas group data are shown in violin plots with the bolded dashed line showing the median and smaller dashed lines showing the upper and lower quartiles for subjects before (blue plot) and after transplant (green plot). (B) Box-and-whisker plots of $CMRO_2$ shown in healthy adults (left; red dots) and in adults with SCD before transplant (middle; blue dots) and 1 year after transplant (right; green dots). $CMRO_2$ does not show significant differences between controls and patients with SCD either before transplant ($P = .18$) or after transplant ($P = .99$).

previous studies have reported stabilization or improvement in cerebral vasculopathy after transplant in children with SCD.[8,44] One area for future study in larger cohorts is whether cerebral vasculopathy improves over time after transplant in adults with SCD.

Here, we extend recent findings providing further evidence that after haplo-BMT, CBF improves to levels observed in healthy age-, race-, and sex-matched adults. These results are similar to CBF findings in 10 children with SCD who underwent HSCT,[23] despite results showing that many adults do not have as much cerebral hemodynamic improvement after blood transfusions as children,[16] indicating that haplo-BMT may provide a more robust change to this hemodynamic measure than transfusion. Furthermore, to the best of our knowledge, this is the first study to show that venous hyperintensity seen on pCASL improved significantly after haplo-BMT, which may indicate reduced capillary level arterio-venous shunting with reduction in highly elevated CBF after transplantation. Theoretically, this reduction in CBF may facilitate improved cerebral oxygen exchange in the brain parenchyma due to healthy capillary transit times and adequate time for oxygen delivery to tissue. These results are consistent with the previous idea that impaired oxygen delivery resulting from arterio-venous shunting contributes to overt strokes and SCIs.[45] Interestingly, a large portion of our participants had donors with sickle cell trait, yet still saw significant hemodynamic improvements in CBF. Prior work by another group has shown that individuals with sickle cell trait (HbAS) have similar CBF and OEF to individuals without sickle cell trait (HbAA); thus, our results are consistent.[46]

Regarding OEF, our findings suggest that there is a large range of OEF levels before transplant, and patients with the highest OEF have reductions in OEF after transplant, or the smallest increases in OEF after transplant when using supplementary models, although there is no significant change in OEF across all participants on average. Furthermore, pretransplant variations in OEF may be because 36% of our sample did not have a cerebrovascular indication for haplo-BMT, in which elevated OEF would be expected. Additionally, 36% of our sample had moyamoya; these participants could not improve oxygen extraction. We investigated differences in transplant-related OEF changes using a cutoff of 1 standard deviation above the mean of our control participants, which has been used in previously published controls (OEF $>$ 42%), although this threshold should be confirmed in additional studies with a larger sample size. Importantly, global OEF was measured via TRUST, and there are several calibration models for converting the blood signal measured on T_2-weighted MRI and hematocrit values into OEF that yield different, albeit generally correlated, results. It should be noted that the only quantitative susceptibility mapping study in HbS and healthy human blood showed no significant difference in relationships between blood water T_2 and Hb type (eg, HbS vs HbA).[47] Here, we chose to use a human HbF model[34] because (1) it was calculated by an established group with experience performing measurements, (2) was calculated over an approximately appropriate anemic and healthy hematocrit range and included a hematocrit dependence, (3) did not abbreviate the blood calibration fitting equation, and (4) provided values consistent with gold-standard PET values in SCD. We did not use a HbS model because it was reported without a Hb dependence (owing to difficulties with manipulating ex vivo red blood cells with HbS without lysing the cells to create a calibration model) and Hb changes significantly before and after transplant. For completeness, we do evaluate our data with all available models in supplemental Material, and the directional changes in OEF are consistent across all models (supplemental Figure 3).

We do not see significant changes in calculated $CMRO_2$ values before and after transplant, and these values are not different from those of healthy controls. A similar finding was reported using ^{15}O PET.[13] This is likely due to compensatory mechanisms in patients with SCD, such as increasing CBF, to allow for oxygen homeostasis. Prior studies using a separate calibration model have shown that in adults with SCD, $CMRO_2$ is significantly reduced,[48,49] so compensation mechanisms may vary with disease severity and related disease complications, such as moyamoya. Conversely, 4 of 11 adults in this cohort are in a less advanced stage of their disease progression, with no cerebrovascular indication for transplant and, therefore, have elevated CBF but have not compensated with elevated OEF as previously described by Juttukonda et al.[16]

The study should also be considered in light of limitations. Although the study includes some of the first clinical and quantitative hemodynamic imaging data from 11 successful adult patients who underwent haplo-BMT, the sample size limits the use of more advanced statistical analyses incorporating a wide range of covariates. Furthermore, although all post–haplo-BMT measures were conducted roughly a year after successful engraftment, the follow-up visits were variable in length (average, 389 ± 150 days), and longer recovery time could result in further improvements in cerebral hemodynamics. Research is needed to understand long-term changes in cerebral hemodynamics measures in both adults and children with SCD after haplo-BMT. Lastly, $CMRO_2$ values were calculated, and this calculation relies on accurate measures of several variables and should be interpreted with care in this and other studies. For these reasons, our primary hypotheses focused on measures of CBF, shunting, and OEF.

In conclusion, we tested the hypothesis that elevated cerebral hemodynamic metrics would normalize to control values after successful haplo-BMT in a large cohort of adults with SCD. After haplo-BMT, adults with SCD had improvement in CBF, with improvements seen in capillary shunting and gray matter values that reduced to values that were not significantly different from control values. Neither OEF nor $CMRO_2$ values were significantly different from healthy control values before and after transplantation. This report adds to the literature on the benefits of HSCT for cerebrovascular health and hemodynamics in adults with SCD.

Acknowledgments

The study was funded by R01 HL155207 (L.C.J. and M.J.D.), K24 HL147017 (L.J.), and R01 NS097763 (M.J.D.) as well as the National Center for Advancing Translational Sciences at Vanderbilt 5UL1TR002243-03. The BMT study had internal funding from Vanderbilt University Medical Center.

Authorship

Contribution: M.A.A. performed research, analyzed data, and wrote the manuscript; W.R. and A.K.S. performed research and analyzed data; L.T.D. analyzed data and provided clinical guidance; S.P. performed research and provided a review of the manuscript; S.D. performed research and was instrumental in data organization;

N.J.P. and C.L. performed research and provided review of the manuscript; A.A.K. recruited the clinical population, provided clinical guidance, and was instrumental in reviewing and preparation the manuscript; M.R.D. provided clinical guidance and was instrumental in reviewing and preparation of the manuscript; M.J.D. designed research, oversaw data analyses, cowrote the manuscript, and provided funding for research project; and L.C.J. designed research, oversaw data analyses, cowrote the manuscript, and provided funding for research project.

Conflict-of-interest disclosure: M.R.D. and his institution sponsor 2 externally funded research investigator-initiated projects; Global Blood Therapeutics (GBT) will provide funding for the cost of the clinical studies but will not be a cosponsor of either study; M.R.D. did not receive any compensation for the conduct of these 2 investigator-initiated observational studies; is a member of the GBT advisory board for a proposed randomized controlled trial for which he receives compensation; is on the steering committee for a Novartis-sponsored phase 2 trial to prevent priapism in men; was a medical adviser in developing the CTX001 Early Economic Model; provided medical input on the economic model as part of an expert reference group for the Vertex/CRISPR CTX001 Early Economic Model in 2020; and consulted for the Formal Pharmaceutical company about SCD in 2021 and 2022. L.C.J. receives minor royalties from UpToDate, an evidence-based clinical information resource, for chapters she has written on stroke in SCD. M.J.D. is a paid consultant for GBT; receives advisory board fee and research-related support from Philips North America; and is the CEO of Biosight LLC, which provides health care technology consulting services. These agreements have been approved by Vanderbilt University Medical Center in accordance with its conflict-of-interest policy. The remaining authors declare no competing financial interests.

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